SORPTION OF AMINO ACIDS AND OTHER SUBSTANCES BY AMBERLITE ION EXCHANGE RESINS*

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MONOBED DEIONIZATION

In these experiments the method of monobed deionization (1), or one step removal of cations and anions, was employed.

Monobed deionization implies a resin bed which contains both acidic groups and basic groups, mixed intimately in definite ratios, and possessing the ability to remove cations and anions simultaneously from solution.

This resinous monobed, adjusted to a neutral pH (7.0), has the capacity to remove all salts from solution. In this manner both acid- and alkali-sensitive systems can be treated for the removal of all ionized constituents.

The potential advantage of such a monobed system, long dreamed of but not an accomplished fact until 1949, is the use of such exchange mixtures in a continuous, countercurrent system whereby a solution is deionized, the resin mixture separated into its component parts, the resin regenerated, remixed, and returned to the treatment cycle continuously.

Thus, in one step, organic solutions may be deionized by contact with both resin exchanger types. The anions are adsorbed after first being converted to the corresponding acids by action of the cation exchangers, after adsorption, the anions are loosely held by the weak base exchanger, and hydrolysis occurs with the resin salt reverting to the free acid. The acidity thus formed builds up rapidly in the resin mixture, and since acid is a regenerant for the cation exchanger, this adsorbent will revert to its original state, and no further ion exchange occurs.†

AMINO ACIDS

It is difficult to determine which substances are selectively removed from complex solutions such as biological fluids after treatment with ion exchange resins. Therefore, to obtain more accurate information on this point, most of our laboratory experiments were confined to studies on solutions of pure substances. However, in some instances, tissue extracts, and human perspiration from normal people, were investigated.

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† In these experiments, Squibb resin Cationex 7, manufactured by the Rohm and Haas Company of Philadelphia, Pa., was used.

Previously, it was shown that choline, lactic acid, and ammonia (2, 3) were absorbed from perspiration by certain exchange resins. These substances exist as anions or cations in solution. In order to determine whether compounds containing both cationic and anionic charges in the same molecule could be removed from solutions, different amino acids were used.

The amino acids which were used, and which are representative of this type of compound, were glycine, a mono-carboxylic-mono-amino acid; lysine, a diamino-monocarboxylic amino acid; glutamic acid, a mono-amino-dicarboxylic amino acid; serine, a hydroxy-amino acid; and arginine, an amino acid containing the guanidino group.

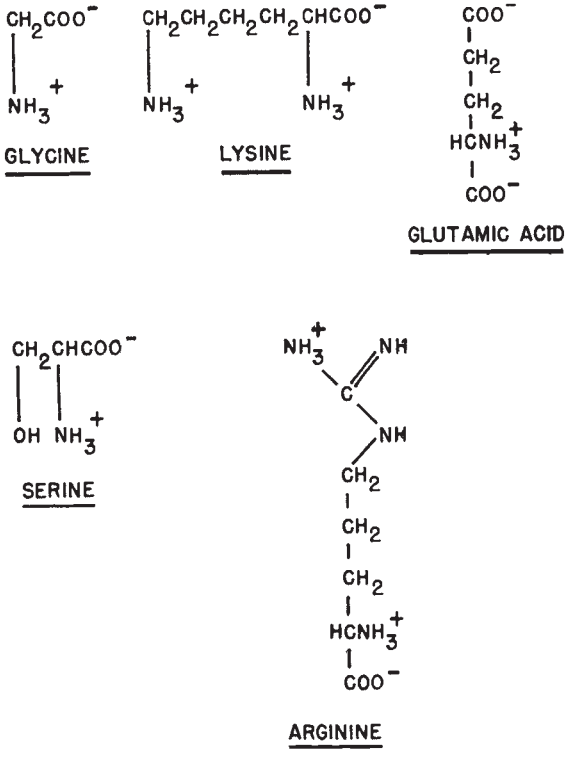

FIG. 1

Figure 1 designates the structural formulae of these five amino acids.

Using the photometric ninhydrin method of Stein and Moore (4), it was found that all of these amino acids were rapidly eliminated from pure solutions.

The fact that these amino acids were absorbed from pure solutions, did not permit conclusions to be drawn concerning their behavior in biological fluids. Therefore, samples of human perspiration from normal people which contained ninhydrin positive substances were treated with the resin. It was found that after stirring the sweat for ten minutes with the resin, no color reaction was obtained

TABLE 1

SORPTION OF AMINO ACIDS BY RESIN CATIONEX 7		
AMINO ACID	α-AMINO N BEFORE RESIN	α-AMINO N AFTER RESIN
	γ	γ
GLYCINE	1400	10
LYSINE	1400	0
ARGININE	1400	0
GLUTAMIC ACID	1400	0
SERINE	1400	0

1 GRAM RESIN + 10 ML. SOLUTION STIRRED MECHANICALLY 15 MINUTES. THE SUSPENSION IS FILTERED AND α-AMINO N DETERMINED ON ALIQUOT ACCORDING TO MOORE AND STEIN.

TABLE 2

SORPTION OF NINHYDRIN POSITIVE SUBSTANCES FROM 1 ML. ECCRINE SWEAT BY RESIN CATIONEX 7

BEFORE RESIN (NINHYDRIN N)	AFTER RESIN (NINHYDRIN N)
100 γ	0 γ

after heating with ninhydrin. It can be concluded, therefore, that the amino acids in human perspiration can be removed completely by sorption on a monobed resin medium.

BACTERIA AND FUNGI

It was desirable to see if the resin monobed could be used to inhibit the growth of bacteria and fungi when measured amounts of the resin were incorporated into culture media.

It was found that the growth of coagulase positive *Staphylococcus aureus* on infusion agar, and of *Monilia albicans* on Sabouraud medium, was inhibited by

specific concentrations of the resin. By varying the resin concentration the culture growth was influenced in varying degree, and eventually growth was completely inhibited. This effect is respectively bacteriostatic and fungistatic, because subcultures of the inhibited organisms, when transferred to fresh media, grew normally.

This inhibition could be caused by at least three possibilities, namely:

1. The resin could remove specific essential metabolites from the media.
2. The resin could lower the salt concentration below physiological limit.
3. The pH of the media could be altered to a degree not compatible with the growth of the organism.

TABLE 3

EFFECT ON GROWTH OF MONILIA ALBICANS ON SABOURAUD MEDIUM WITH RESIN CATIONEX 7.

RESIN CONCENTRATION	GROWTH OF FUNGI	pH
10.0 %	VERY SLIGHTLY	6.0
12.5 %	0	6.0
15.0 %	0	6.0
CONTROL MONILIA ALBICANS ON SABOURAUD WITHOUT RESIN:	+ + +	6.0-7.0

TABLE 4

GROWTH OF STAPH. AUREUS ON PEPTONE AGAR WITH RESIN CATIONEX 7.

RESIN CONCENTRATION	GROWTH OF STAPHYLOCOCCUS AUREUS	pH
10.0%	+ +	6.0
12.5%	+	5.0
15.0%	+	5.0
CONTROL OF STAPHYLOCOCCUS AUREUS WITHOUT RESIN:	+ + +	8.0

TABLE 5

EFFECT OF VARYING CONCENTRATIONS OF RESIN CATIONEX 7 IN SABOURAUD MEDIUM ON GROWTH OF MONILIA ALBICANS

AMBERLITE CONC.	GROWTH OF MONILIA	pH
1%	+ + +	6.5
4%	+ + +	6.0
7%	+ + +	5.0
10%	+ +	5.0
12.5%	NEGATIVE	6.0
15%	0	6.0
17.5%	0	5.5
20%	0	5.0

It is believed that the third possibility may be excluded since only minor changes in the pH were observed.

ALLERGENIC SUBSTANCES

It was interesting to observe that, in addition to the removal of certain substances necessary for the growth of bacteria and fungi, that the resin monobed absorbed compounds, presumably toxins, elaborated by these organisms.

For example, it was found that a commercial allergenic extract,* which produced marked local skin reactions when 0.1 cc. was injected intradermally in five human subjects, was rendered ineffective after filtration through the resin monobed. It is probable that the specific allergenic substance in this preparation was removed by the resin.

ARGINASE

In our experiments with arginase so much difficulty was encountered that as yet this work is not suitable for publications; but, we are able to confirm the fact that no arginase is to be found in human perspiration.

The potential efficacy of exchange resins in clinical dermatology is based on the ability of the resin exchangers to remove a great variety of ionic substances from both simple and complex solutions. We believe that these laboratory ob-

* This allergenic extract was Special Fungus Mixture #4 (Trichophyton gypseum, Trichophyton interdigital, Monilia albicans, Epidermophyton inguinale), 1 to 5000 solution, Arlington Chemical Co., Lot No. 5370.

TABLE 6

TREATMENT OF ALLERGENIC EXTRACT # 4* WITH SQUIBB RESIN CATIONEX 7.

100 MG. RESIN WASHED WITH 5 CC. H_2O AND SUCKED THROUGH A SEITZ FILTER. THEN 1 CC. OF ALLERGENIC EXTRACT WAS PASSED THROUGH THE WASHED RESIN (STERILE).

INTRACUTANEOUS TEST WITH ALLERGENIC EXTRACT

PATIENT	READINGS: HOURS	ALLERGENIC EXTRACT	FILTERED ALLERGENIC EXTRACT	CONTROL: BUFFERED SALINE
J.M.	24	+ + + + 4 CM. DIA., RED, INFILTRATED, IRRITATIVE WHEAL	0	0
	48	+ + + 3.5 CM. DIA. IRRITATIVE ERYTHMMATOUS WHEAL	0	0
	72	+ + 2 CM. DIA. RED INFILTRATED PAPULE	0	0
	168	+ FLAT ITCHING ERYTHEMA WHEALS WITH FRICTION	0	0
	240	4 CM. DIA. MACULE PALE BROWN STAINING	0	0

* THE ALLERGENIC EXTRACT WAS SPECIAL FUNGUS MIXTURE # 4 (TRICHOPHYTON GYPSEUM, TRICHOPHYTON INTERDIGITAL, MONILIA ALBICANS, EPIDERMOPHYTON INGUINALE, 1 TO 5000 SOLUTION. ARLINGTON CHEMICAL CO., LOT NO. 5370.

servations are sound, and that they should encourage clinical investigation of the application of monobed deionization as a topical therapy.

SUMMARY

1. The process of monobed deionization, or the simultaneous removal of cations and anions from solutions, is described.

2. Ion exchange resins have the sorption capacity to remove amino acids *in vitro*, and the amino acids present in human perspiration.

3. Ion exchange resins were found to be bacteriostatic for *Staphylococcus aureus*, and fungistatic for *Monilia albicans* in direct proportion to the concentration of the resin in culture media.

4. The allergenic substance contained in a fungous extract derived from four different pathogenic fungi was rendered innocuous for intradermal skin testing in people whom previously had reacted positively—after filtering the extract in a Seitz filter containing the resin.

5. No arginase was found in human perspiration.

REFERENCES

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ANNOUNCEMENT

The American Academy of Dermatology and Syphilology revised its Constitution at the recent Academy meeting in December, 1953, to permit membership of physicians living outside the United States. The new Section 3 of Article III of the Constitution reads as follows:

"Any physician in good standing as a teacher or in research work or with three years full-time experience in the practice of dermatology and syphilology in any country other than the United States shall be eligible to apply for Non-Resident Fellowship. The Committee on Membership shall, if necessary, appoint a special committee to investigate any applicant. Requirements for Non-Resident Fellowship shall be approximately equivalent to the requirements for certification by the American Board of Dermatology and Syphilology".

The Constitution further states that "Non-Resident Fellows . . . shall be entitled to all privileges of Fellowship except serving as officers, directors or members of committees".

The American Academy of Dermatology and Syphilology invites qualified dermatologists in countries other than the United States who are interested in becoming Non-Resident Fellows to write to John E. Rauschkolb, M.D., Secretary, Box 6565, Cleveland 1 Ohio, for a membership application.